

SEQUENCE LISTING

<110> Bron, Sierd Jongbloed, Jan D.H. Mueller, Joerg P. Van Dijl, Jan M. <120> Twin-Arginine Translocation in Bacillus <130> GC634-2 <140> US 09/954,737 <141> 2001-09-17 <150> US 60/233,610 <151> 2000-09-18 <160> 83 <170> FastSEQ for Windows Version 4.0 <210> 1 <211> 89 <212> PRT <213> Escherichia coli <400> 1 Met Gly Gly Ile Ser Ile Trp Gln Leu Leu Ile Ile Ala Val Ile Val 10 Val Leu Leu Phe Gly Thr Lys Lys Leu Gly Ser Ile Gly Ser Asp Leu 25 Gly Ala Ser Ile Lys Gly Phe Lys Lys Ala Met Ser Asp Asp Glu Pro 40 Lys Gln Asp Lys Thr Ser Gln Asp Ala Asp Phe Thr Ala Lys Thr Ile 55 60 Ala Asp Lys Gln Ala Asp Thr Asn Gln Glu Gln Ala Lys Thr Glu Asp 70 80 Ala Lys Arg His Asp Lys Glu Gln Val <210> 2 <211> 67 <212> PRT <213> Escherichia coli <400> 2 Met Gly Glu Ile Ser Ile Thr Lys Leu Leu Val Val Ala Ala Leu Val

```
His Lys Glu
65
<210> 3
<211> 57
<212> PRT
<213> Bacillus subtilis
<400> 3
Met Pro Ile Gly Pro Gly Ser Leu Ala Val Ile Ala Ile Val Ala Leu
                                    10
Ile Ile Phe Gly Pro Lys Lys Leu Pro Glu Leu Gly Lys Ala Ala Gly
                               25
Asp Thr Leu Arg Glu Phe Lys Asn Ala Thr Lys Gly Leu Thr Ser Asp
                            40
Glu Glu Glu Lys Lys Lys Glu Asp Gln
<210> 4
<211> 70
<212> PRT
<213> Bacillus subtilis
<400> 4
Met Phe Ser Asn Ile Gly Ile Pro Gly Leu Ile Leu Ile Phe Val Ile
                                    10
Ala Ile Ile Ile Phe Gly Pro Ser Lys Leu Pro Glu Ile Gly Arg Ala
                                25
Ala Lys Arg Thr Leu Leu Glu Phe Lys Ser Ala Thr Lys Ser Leu Val
Ser Gly Asp Glu Lys Glu Glu Lys Ser Ala Glu Leu Thr Ala Val Lys
                        55
Gln Asp Lys Asn Ala Gly
<210> 5
<211> 62
<212> PRT
<213> Bacillus subtilis
<400> 5
Met Glu Leu Ser Phe Thr Lys Ile Leu Val Ile Leu Phe Val Gly Phe
                 5
Leu Val Phe Gly Pro Asp Lys Leu Pro Ala Leu Gly Arg Ala Ala Gly
                                25
Lys Ala Leu Ser Glu Phe Lys Gln Ala Thr Ser Gly Leu Thr Gln Asp
                           40
Ile Arg Lys Asn Asp Ser Glu Asn Lys Glu Asp Lys Gln Met
                       55
<210> 6
<211> 171
<212> PRT
<213> Escherichia coli
<400> 6
Met Phe Asp Ile Gly Phe Ser Glu Leu Leu Leu Val Phe Ile Ile Gly
```

```
5
Leu Val Val Leu Gly Pro Gln Arg Leu Pro Val Ala Val Lys Thr Val
Ala Gly Trp Ile Arg Ala Leu Arg Ser Leu Ala Thr Thr Val Gln Asn
                           40
Glu Leu Thr Gln Glu Leu Lys Leu Gln Glu Phe Gln Asp Ser Leu Lys
                       55
Lys Val Glu Lys Ala Ser Leu Thr Asn Leu Thr Pro Glu Leu Lys Ala
                                       75
                   70
Ser Met Asp Glu Leu Arg Gln Ala Ala Glu Ser Met Lys Arg Ser Tyr
                                   90
               8.5
Val Ala Asn Asp Pro Glu Lys Ala Ser Asp Glu Ala His Thr Ile His
                               105
Asn Pro Val Val Lys Asp Asn Glu Ala Ala His Glu Gly Val Thr Pro
                                               125
                           120
Ala Ala Gln Thr Gln Ala Ser Pro Glu Gln Lys Pro Glu Thr
                       135
Thr Pro Glu Pro Val Val Lys Pro Ala Ala Asp Ala Glu Pro Lys Thr
                   150
                                      155
Ala Ala Pro Ser Pro Ser Ser Ser Asp Lys Pro
               165
<210> 7
<211> 258
<212> PRT
<213> Escherichia coli
<400> 7
Met Ser Val Glu Asp Thr Gln Pro Leu Ile Thr His Leu Ile Glu Leu
Arg Lys Arg Leu Leu Asn Cys Ile Ile Ala Val Ile Val Ile Phe Leu
                               25
Cys Leu Val Tyr Phe Ala Asn Asp Ile Tyr His Leu Val Ser Ala Pro
                           40
Leu Ile Lys Gln Leu Pro Gln Gly Ser Thr Met Ile Ala Thr Asp Val
                       55
Ala Ser Pro Phe Phe Thr Pro Ile Lys Leu Thr Phe Met Val Ser Leu
                                       75
                   70
Ile Leu Ser Ala Pro Val Ile Leu Tyr Gln Val Trp Ala Phe Ile Ala
               85
                                   90
Pro Ala Leu Tyr Lys His Glu Arg Arg Leu Val Val Pro Leu Leu Val
                               105
           100
Ser Ser Ser Leu Leu Phe Tyr Ile Gly Met Ala Phe Ala Tyr Phe Val
                           120
Val Phe Pro Leu Ala Phe Gly Phe Leu Ala Asn Thr Ala Pro Glu Gly
                       135
                                           140
Val Gln Val Ser Thr Asp Ile Ala Ser Tyr Leu Ser Phe Val Met Ala
                                       155
                   150
Leu Phe Met Ala Phe Gly Val Ser Phe Glu Val Pro Val Ala Ile Val
                                   170
               165
Leu Leu Cys Trp Met Gly Ile Thr Ser Pro Glu Asp Leu Arg Lys Lys
                               185
Arg Pro Tyr Val Leu Val Gly Ala Phe Val Val Gly Met Leu Leu Thr
                           200
        195
Pro Pro Asp Val Phe Ser Gln Thr Leu Leu Ala Ile Pro Met Tyr Cys
                        215
Leu Phe Glu Ile Gly Val Phe Phe Ser Arg Phe Tyr Val Gly Lys Gly
```

```
Arg Asn Arg Glu Glu Glu Asn Asp Ala Glu Ala Glu Ser Glu Lys Thr
               245
                               250
Glu Glu
<210> 8
<211> 254
<212> PRT
<213> Bacillus subtilis
<400> 8
Met Thr Arg Met Lys Val Asn Gln Met Ser Leu Leu Glu His Ile Ala
Glu Leu Arg Lys Arg Leu Leu Ile Val Ala Leu Ala Phe Val Val Phe
                               25
           20
Phe Ile Ala Gly Phe Phe Leu Ala Lys Pro Ile Ile Val Tyr Leu Gln
Glu Thr Asp Glu Ala Lys Gln Leu Thr Leu Asn Ala Phe Asn Leu Thr
                      55
                                         60
Asp Pro Leu Tyr Val Phe Met Gln Phe Ala Phe Ile Ile Gly Ile Val
                                      75
                   70
Leu Thr Ser Pro Val Ile Leu Tyr Gln Leu Trp Ala Phe Val Ser Pro
                                  90
              85
Gly Leu Tyr Glu Lys Glu Arg Lys Val Thr Leu Ser Tyr Ile Pro Val
                              105
           100
Ser Ile Leu Leu Phe Leu Ala Gly Leu Ser Phe Ser Tyr Tyr Ile Leu
                          120
Phe Pro Phe Val Val Asp Phe Met Lys Arg Ile Ser Gln Asp Leu Asn
                               140
                       135
Val Asn Gln Val Ile Gly Ile Asn Glu Tyr Phe His Phe Leu Leu Gln
                                      155
                   150
Leu Thr Ile Pro Phe Gly Leu Leu Phe Gln Met Pro Val Ile Leu Met
                                 170
Phe Leu Thr Arg Leu Gly Ile Val Thr Pro Met Phe Leu Ala Lys Ile
                              185
          180
Arg Lys Tyr Ala Tyr Phe Thr Leu Leu Val Ile Ala Ala Leu Ile Thr
      195
                          200
Pro Pro Glu Leu Leu Ser His Met Met Val Thr Val Pro Leu Leu Ile
                      215
                                         220
Leu Tyr Glu Ile Ser Ile Leu Ile Ser Lys Ala Ala Tyr Arg Lys Ala
                  230
Gln Lys Ser Ser Ala Ala Asp Arg Asp Val Ser Ser Gly Gln
<210> 9
<211> 245
<212> PRT
<213> Bacillus subtilis
<400> 9
Met Asp Lys Lys Glu Thr His Leu Ile Gly His Leu Glu Glu Leu Arg
                                  10
               5
Arg Arg Ile Ile Val Thr Leu Ala Ala Phe Phe Leu Phe Leu Ile Thr
                               25
Ala Phe Leu Phe Val Gln Asp Ile Tyr Asp Trp Leu Ile Arg Asp Leu
                           40
Asp Gly Lys Leu Ala Val Leu Gly Pro Ser Glu Ile Leu Trp Val Tyr
```

```
55
Met Met Leu Ser Gly Ile Cys Ala Ile Ala Ala Ser Ile Pro Val Ala
                                       75
                   70
Ala Tyr Gln Leu Trp Arg Phe Val Ala Pro Ala Leu Thr Lys Thr Glu
              85
                                   90
Arg Lys Val Thr Ile Met Tyr Ile Met Tyr Ile Pro Gly Leu Phe Ala
                               105
Leu Phe Leu Ala Gly Ile Ser Phe Gly Tyr Phe Val Leu Phe Pro Ile
                                               125
                           120
Val Leu Ser Phe Leu Thr His Leu Ser Ser Gly His Phe Glu Thr Met
                      135
                                          140
Phe Thr Ala Asp Arg Tyr Phe Arg Phe Met Val Asn Leu Ser Leu Pro
                                      155
                   150
Phe Gly Phe Leu Phe Glu Met Pro Leu Val Val Met Phe Leu Thr Arg
                                   170
               165
Leu Gly Ile Leu Asn Pro Tyr Arg Leu Ala Lys Ala Arg Lys Leu Ser
                               185
           180
Tyr Phe Leu Leu Ile Val Val Ser Ile Leu Ile Thr Pro Pro Asp Phe
                           200
Ile Ser Asp Phe Leu Val Met Ile Pro Leu Leu Val Leu Phe Glu Val
                                           220
                       215
Ser Val Thr Leu Ser Ala Phe Val Tyr Lys Lys Arg Met Arg Glu Glu
                                       235
                  230
Thr Ala Ala Ala Ala
               245
<210> 10
<211> 63
<212> PRT
<213> Bacillus alcalophilus
<400> 10
Met Gly Gly Leu Ser Val Gly Ser Val Val Leu Ile Ala Leu Val Ala
                                   10
Leu Leu Ile Phe Gly Pro Lys Lys Leu Pro Glu Leu Gly Lys Ala Ala
                               25
Gly Ser Thr Leu Arg Glu Phe Lys Asn Ala Thr Lys Gly Leu Ala Asp
                           40
Asp Asp Asp Asp Thr Lys Ser Thr Asn Val Gln Lys Glu Lys Ala
   50
<210> 11
<211> 272
<212> PRT
<213> Bacillus alcalophilus
<400> 11
Met Thr Met Met Thr Pro Asn Gln Gln Thr Ser Lys Lys Lys Arg
                                   10
                5
Lys Gly Arg Lys Gly Arg Val Pro Met Gln Asp Met Ser Ile Met Asp
                               25
           20
His Ala Glu Glu Leu Arg Arg Ile Phe Val Val Leu Ala Phe Phe
                            40
Ile Val Ala Leu Ile Gly Gly Phe Phe Leu Ala Val Pro Val Ile Thr
                                           60
                        55
Phe Leu Gln Asn Ser Pro Gln Ala Ala Asp Met Pro Phe Asn Ala Phe
                                        75
```

```
Arg Leu Thr Asp Pro Leu Arg Val Tyr Met Asn Phe Ala Val Ile Thr
                                   90
               85
Ala Leu Val Leu Ile Ile Pro Val Ile Leu Tyr Gln Leu Trp Ala Phe
           100
                               105
Val Ser Pro Gly Leu Lys Glu Asn Glu Gln Lys Ala Thr Leu Ala Tyr
                          120
Ile Pro Ile Ala Phe Leu Leu Phe Leu Ala Gly Ile Ala Phe Ser Tyr
                       135
                                          140
Phe Ile Leu Pro Phe Val Ile Ser Phe Met Gly Gln Met Ala Asp
                   150
                                       155
Arg Leu Glu Ile Asn Glu Met Tyr Gly Ile Asn Glu Tyr Phe Ser Phe
                                   170
               165
Leu Phe Gln Leu Thr Ile Pro Phe Gly Leu Leu Phe Gln Leu Pro Val
                               185
           180
Val Val Met Phe Leu Thr Arg Leu Gly Val Val Thr Pro Thr Phe Leu
                           200
                                               205
Arg Lys Ile Arg Lys Tyr Ala Tyr Phe Ala Leu Leu Val Ile Ala Gly
                                           220
                      215
Ile Ile Thr Pro Pro Glu Leu Thr Ser His Leu Phe Val Thr Val Pro
                                      235
                   230
Met Leu Ile Leu Tyr Glu Ile Ser Ile Thr Ile Ser Ala Ile Thr Tyr
                           250
            245
Arg Lys Tyr His Gly Thr Thr Asp His Asn Gly Gln Glu Ser Ala Lys
                               265
<210> 12
<211> 35
<212> DNA
<213> Artificial Sequence
<220>
<223> primer
<400> 12
                                                                       35
cccaagetta tgaaagggag ggettttttg aatgg
<210> 13
<211> 26
<212> DNA
<213> Artificial Sequence
<220>
<223> primer
<400> 13
                                                                       26
gcggatccaa agctgagcac gatcgg
<210> 14
<211> 39
<212> DNA
<213> Artificial Sequence
<220>
<223> primer
<400> 14
                                                                       39
cccaagctta aaaagaaaga agatcagtaa gttaggatg
```

<210> <211> <212> <213>	25	
<220> <223>	primer	
<400> gcggat		25
<210> <211> <212> <213>	21	
<220> <223>	primer	
<400> ggaatt		21
<210> <211> <212> <213>	21	
<220> <223>	primer	
<400> cgggat	17 cccat catgggaagc g	21
<210> <211> <212> <213>	26	
<220> <223>	primer	
<400> ggggta	18 accgg aaaacgcttg atcagg	26
<210> <211> <212> <213>	22	
<220> <223>	primer	
<400> cggga	19 tcctt tgggcgatag cc	22
Z210N	20	

<211> 42	
<212> DNA	
<213> Artificial Sequence	
Allo, McClical Sequence	
1000	
<220>	
<223> primer	
<400> 20	
gaggatccat gaggagagag gggatcttga atggcatacg ac	42
gaggattotae gaggagagag gggattottga at gggttottga at	
(210) 21	
<210> 21	
<211> 27	
<212> DNA	
<213> Artificial Sequence	
<220>	
<223> primer	
12237 primer	
<400> 21	07
cgatcctgca ggacctcatc ggattgc	27
<210> 22	
<211> 27	
<212> DNA	
<213> Artificial Sequence	
<220>	
<223> primer	
- Pariss	
<400> 22	
	27
gtaggatccg cgcctaactt ctcaagc	2,
<210> 23	
<211> 25	
<212> DNA	
<213> Artificial Sequence	
Allo, metiletal soquenes	
1000	
<220>	
<223> primer	
<400> 23	
atagaattca aaaaggaaga gtatg	25
35 5 5 5	
<210> 24	
<211> 24	
<212> DNA	
<213> Artificial Sequence	
<220>	
<223> primer	
CEO. PIIMOE	
(100) 24	
<400> 24	24
ctggggatcc aaaaacagga aggc	44
<210> 25	
<211> 35	
<212> DNA	

<213>	Artificial Sequence		
<220>			
	primer		
44005			
<400>	ggtog acgcagcatt tacttcaaag gcccc	35	
yayaaç	gytty atytatt tatteaung gette		
<210>	26		
<211>			
<212>			
<213>	Artificial Sequence		
<220>			
<223>	primer		
<400>		26	
accggg	gtcga ccgtcgtttt acaacg		
<210>	27		
<211>			
<212>			
<213>	Artificial Sequence		
<220>			
<223>	primer		
400			
<400>	2/ ctcat ggcctgcccg gtt	23	
gggaat	cical gycolycoly yel		
<210>	28		
<211>			
<212>			
<213>	Artificial Sequence		
<220>			
<223>	primer		
. 4 0 0 .			
<400>	28 atccc gaattaagga gtgg	24	
caayyo	accec gaaccaagga gegg		
<210>	29		
<211>			
<212>			
<213>	Artificial Sequence		
<220>			
	primer		
<400>		27	
ggtctgcagc tgcactaagc ggccgcc 27			
<210>	30		
<211>			
<212>			
<213>	Artificial Sequence		

```
<220>
<223> predicted twin-arginine (RR-)signal peptides of B.
      subtilis
<400> 30
Arg Arg Ile Leu Leu
<210> 31
<211> 5
<212> PRT
<213> Artificial Sequence
<223> predicted twin-arginine (RR-)signal peptides of B.
      subtilis
<400> 31
Arg Arg Ser Phe Glu
<210> 32
<211> 5
<212> PRT
<213> Artificial Sequence
<223> predicted twin-arginine (RR-)signal peptides of B.
      subtilis
<400> 32
Arg Arg Thr Leu Met
<210> 33
<211> 5
<212> PRT
<213> Artificial Sequence
<223> predicted twin-arginine (RR-)signal peptides of B.
      subtilis
<400> 33
Arg Arg Ile Ile Ala
<210> 34
<211> 5
<212> PRT
<213> Artificial Sequence
<223> predicted twin-arginine (RR-)signal peptides of B.
      subtilis
<400> 34
```

```
Arg Arg Leu Val Tyr
1
<210> 35
<211> 5
<212> PRT
<213> Artificial Sequence
<223> predicted twin-arginine (RR-)signal peptides of B.
      subtilis
<400> 35
Arg Arg Lys Leu
                 5
1
<210> 36
<211> 5
<212> PRT
<213> Artificial Sequence
<223> predicted twin-arginine (RR-) signal peptides of B.
     subtilis
<400> 36
Arg Arg Lys Phe Ile
                 5
1
<210> 37
<211> 5
<212> PRT
<213> Artificial Sequence
<223> predicted twin-arginine (RR-)signal peptides of B.
      subtilis
<400> 37
Arg Arg Gln Phe Leu
1
                5
<210> 38
<211> 5
<212> PRT
<213> Artificial Sequence
<223> predicted twin-arginine (RR-)signal peptides of B.
      subtilis
<400> 38
Arg Arg Leu Ile Ile
                 5
1
<210> 39
<211> 5
```

```
<212> PRT
<213> Artificial Sequence
<220>
<223> predicted twin-arginine (RR-)signal peptides of B.
      subtilis
<400> 39
Arg Arg Asn Phe Lys
<210> 40
<211> 5
<212> PRT
<213> Artificial Sequence
<220>
<223> predicted twin-arginine (RR-)signal peptides of B.
     subtilis
<400> 40
Arg Arg Lys Phe Ser
<210> 41
<211> 5
<212> PRT
<213> Artificial Sequence
<220>
<223> predicted twin-arginine (RR-)signal peptides of B.
      subtilis
<400> 41
Arg Arg Ala Phe Leu
<210> 42
<211> 5
<212> PRT
<213> Artificial Sequence
<220>
<223> predicted twin-arginine (RR-)signal peptides of B.
      subtilis
<400> 42
Arg Arg Met Lys Ile
<210> 43
<211> 5
<212> PRT
<213> Artificial Sequence
<220>
<223> predicted twin-arginine (RR-) signal peptides of B.
```

subtilis <400> 43 Arg Arg Ser Cys Leu <210> 44 <211> 5 <212> PRT <213> Artificial Sequence <220> <223> predicted twin-arginine (RR-)signal peptides of B. subtilis <400> 44 Arg Arg Thr His Val <210> 45 <211> 5 <212> PRT <213> Artificial Sequence <223> predicted twin-arginine (RR-)signal peptides of B. subtilis <400> 45 Arg Arg Val Ala Ile <210> 46 <211> 5 <212> PRT <213> Artificial Sequence <223> predicted twin-arginine (RR-)signal peptides of B. subtilis <400> 46 Arg Arg Gln Phe Leu <210> 47 <211> 5 <212> PRT <213> Artificial Sequence <223> predicted twin-arginine (RR-)signal peptides of B. subtilis

<400> 47

1

Arg Arg Phe Leu Leu

```
<210> 48
<211> 5
<212> PRT
<213> Artificial Sequence
<223> predicted twin-arginine (RR-)signal peptides of B.
      subtilis
<400> 48
Arg Arg Asn Thr Val
1
<210> 49
<211> 5
<212> PRT
<213> Artificial Sequence
<223> predicted twin-arginine (RR-) signal peptides of B.
      subtilis
<400> 49
Arg Arg Leu Leu Met
                 5
1
<210> 50
<211> 5
<212> PRT
<213> Artificial Sequence
<223> predicted twin-arginine (RR-)signal peptides of B.
      subtilis
<400> 50
Arg Arg Lys Ile Leu
 1
                 5
<210> 51
<211> 5
<212> PRT
<213> Artificial Sequence
<223> predicted twin-arginine (RR-)signal peptides of B.
      subtilis
<400> 51
Arg Arg Asp Ile Leu
                 5
 1
<210> 52
<211> 42
<212> PRT
<213> Artificial Sequence
```

```
<220>
<223> predicted twin-arginine signal peptides of B.
      subtilis
<400> 52
Ser Pro Ala Gln Arg Arg Ile Leu Leu Tyr Ile Leu Ser Phe Ile Phe
                        10
        5
Val Ile Gly Ala Val Val Tyr Phe Val Lys Ser Asp Tyr Leu Phe Thr
Leu Ile Phe Ile Ala Ile Ala Ile Leu Phe
                           40
<210> 53
<211> 30
<212> PRT
<213> Artificial Sequence
<220>
<223> predicted twin-arginine signal peptides of B.
     subtilis
<400> 53
Met Val Ser Ile Arg Arg Ser Phe Glu Ala Tyr Val Asp Asp Met Asn
               5
                                   10
Ile Ile Thr Val Leu Ile Pro Ala Glu Gln Lys Glu Ile Met
            20
                               25
<210> 54
<211> 32
<212> PRT
<213> Artificial Sequence
<220>
<223> predicted twin-arginine signal peptides of B.
      subtilis
<400> 54
Met Ala Ala Tyr Ile Ile Arg Arg Thr Leu Met Ser Ile Pro Ile Leu
                                   10
Leu Gly Ile Thr Ile Leu Ser Phe Val Ile Met Lys Ala Ala Pro Gly
            20
                                25
<210> 55
<211> 34
<212> PRT
<213> Artificial Sequence
<223> predicted twin-arginine signal peptides of B.
      subtilis
<400> 55
Met Lys Phe Val Lys Arg Arg Ile Ile Ala Leu Val Thr Ile Leu Met
                                    10
Leu Ser Val Thr Ser Leu Phe Ala Leu Gln Pro Ser Ala Lys Ala Ala
```

```
Glu His
<210> 56
<211> 32
<212> PRT
<213> Artificial Sequence
<220>
<223> predicted twin-arginine signal peptides of B.
<400> 56
Met Leu Lys Tyr Ile Gly Arg Arg Leu Val Tyr Met Ile Ile Thr Leu
Phe Val Ile Val Thr Val Thr Phe Phe Leu Met Gln Ala Ala Pro Gly
            20
                                25
<210> 57
<211> 42
<212> PRT
<213> Artificial Sequence
<220>
<223> predicted twin-arginine signal peptides of B.
      subtilis
<400> 57
Met Thr Ser Pro Thr Arg Arg Arg Thr Ala Lys Arg Arg Arg Lys
Leu Asn Lys Arg Gly Lys Leu Leu Phe Gly Leu Leu Ala Val Met Val
Cys Ile Thr Ile Trp Asn Ala Leu His Arg
                            40
<210> 58
<211> 54
<212> PRT
<213> Artificial Sequence
<223> predicted twin-arginine signal peptides of B.
      subtilis
<400> 58
Met Ala Tyr Asp Ser Arg Phe Asp Glu Trp Val Gln Lys Leu Lys Glu
Glu Ser Phe Gln Asn Asn Thr Phe Asp Arg Arg Lys Phe Ile Gln Gly
                                25
Ala Gly Lys Ile Ala Gly Leu Ser Leu Gly Leu Thr Ile Ala Gln Ser
                            40
        35
Val Gly Ala Phe Glu Val
    50
<210> 59
<211> 36
<212> PRT
```

<213> Artificial Sequence

```
<220>
<223> predicted twin-arginine signal peptides of B.
      subtilis
<400> 59
Met Gly Gly Lys His Asp Ile Ser Arg Arg Gln Phe Leu Asn Tyr Thr
               5
                            10
Leu Thr Gly Val Gly Gly Phe Met Ala Ala Ser Met Leu Met Pro Met
                               25
Val Arg Phe Ala
       35
<210> 60
<211> 26
<212> PRT
<213> Artificial Sequence
<220>
<223> predicted twin-arginine signal peptides of B.
     subtilis
<400> 60
Met Leu Leu Lys Arg Arg Ile Gly Leu Leu Leu Ser Met Val Gly Val
                                    10
               5
Phe Met Leu Leu Ala Gly Cys Ser Ser Val
<210> 61
<211> 39
<212> PRT
<213> Artificial Sequence
<220>
<223> predicted twin-arginine signal peptides of B.
      subtilis
<400> 61
Met Lys Lys Thr Leu Thr Thr Ile Arg Arg Ser Ser Ile Ala Arg Arg
                                    10
Leu Ile Ile Ser Phe Leu Leu Ile Leu Ile Val Pro Ile Thr Ala Leu
            20
                                25
Ser Val Ser Ala Tyr Gln Ser
        35
<210> 62
<211> 35
<212> PRT
<213> Artificial Sequence
<223> predicted twin-arginine signal peptides of B.
     subtilis
<400> 62
Met Lys Lys Arg Lys Arg Arg Asn Phe Lys Arg Phe Ile Ala Ala Phe
```

```
Leu Val Leu Ala Leu Met Ile Ser Leu Val Pro Ala Asp Val Leu Ala
                                25
            20
Lys Ser Thr
       35
<210> 63
<211> 33
<212> PRT
<213> Artificial Sequence
<220>
<223> predicted twin-arginine signal peptides of B.
      subtilis
<400> 63
Lys Arg Arg Lys Phe Ser Ser Val Val Ala Ala Val Leu Ile Phe Ala
                                    10
1
Leu Ile Phe Ser Leu Phe Ser Pro Gly Thr Lys Ala Ala Ala Gly
                                25
Ala
<210> 64
<211> 35
<212> PRT
<213> Artificial Sequence
<223> predicted twin-arginine signal peptides of B.
      subtilis
<400> 64
Met Glu Met Phe Asp Leu Glu Phe Met Arg Arg Ala Phe Leu Ala Gly
                                   10
Gly Met Ile Ala Val Met Ala Pro Ile Leu Gly Val Tyr Leu Val Leu
                                25
Arg Arg Gln
       35
<210> 65
<211> 26
<212> PRT
<213> Artificial Sequence
<223> predicted twin-arginine signal peptides of B.
     subtilis
Met Lys Lys Arg Arg Lys Ile Cys Tyr Cys Asn Thr Ala Leu Leu
Met Ile Leu Leu Ala Gly Cys Thr Asp Ser
            20
<210> 66
<211> 20
<212> PRT
<213> Artificial Sequence
```

```
<220>
<223> predicted twin-arginine signal peptides of B.
      subtilis
<400> 66
Met Arg Arg Ile Leu Ser Ile Leu Val Phe Ala Ile Met Leu Ala Gly
Cys Ser Ser Asn
<210> 67
<211> 43
<212> PRT
<213> Artificial Sequence
<220>
<223> predicted twin-arginine signal peptides of B.
      subtilis
<400> 67
Met Ser Ala Gly Lys Ser Tyr Arg Lys Lys Met Lys Gln Arg Arg Met
                                    10
Asn Met Lys Ile Ser Lys Tyr Ala Leu Gly Ile Leu Met Leu Ser Leu
                                25
Val Phe Val Leu Ser Ala Cys Gly Asn Asn Asn
                            40
<210> 68
<211> 42
<212> PRT
<213> Artificial Sequence
<220>
<223> predicted twin-arginine signal peptides of B.
      subtilis
<400> 68
Lys Lys Arg Val Ala Gly Trp Tyr Arg Arg Met Lys Ile Lys Asp Lys
1
Leu Phe Val Phe Leu Ser Leu Ile Met Ala Val Ser Phe Leu Phe Val
Tyr Ser Gly Val Gln Tyr Ala Phe His Val
<210> 69
<211> 38
<212> PRT
<213> Artificial Sequence
<223> predicted twin-arginine signal peptides of B.
     subtilis
<400> 69
Met Arg Arg Ser Cys Leu Met Ile Arg Arg Arg Lys Arg Met Phe Thr
```

```
Ala Val Thr Leu Leu Val Leu Leu Val Met Gly Thr Ser Val Cys Pro
            20
Val Lys Ala Glu Gly Ala
        35
<210> 70
<211> 38
<212> PRT
<213> Artificial Sequence
<220>
<223> predicted twin-arginine signal peptides of B.
      subtilis
<400> 70
Met Arg Ile Gln Lys Arg Arg Thr His Val Glu Asn Ile Leu Arg Ile
                 5
Leu Leu Pro Pro Ile Met Ile Leu Ser Leu Ile Leu Pro Thr Pro Pro
                                25
Ile His Ala Glu Glu Ser
       35
<210> 71
<211> 32
<212> PRT
<213> Artificial Sequence
<220>
<223> predicted twin-arginine signal peptides of B.
      subtilis
<400> 71
Met Leu Arg Asp Leu Gly Arg Arg Val Val Ala Ile Ala Ala Ile Leu
                                   10
Ser Gly Ile Ile Leu Gly Gly Met Ser Ile Ser Leu Ala Asn Met Pro
                                25
<210> 72
<211> 34
<212> PRT
<213> Artificial Sequence
<220>
<223> predicted twin-arginine signal peptides of B.
      subtilis
<400> 72
Met Lys Lys Met Ser Arg Arg Gln Phe Leu Lys Gly Met Phe Gly Ala
                5
                                    10
Leu Ala Ala Gly Ala Leu Thr Ala Gly Gly Gly Tyr Gly Tyr Ala Arg
                                25
            20
Tyr Leu
<210> 73
<211> 28
<212> PRT
```

<213> Artificial Sequence

```
<220>
<223> predicted twin-arginine signal peptides of B.
      subtilis
<400> 73
Met Arg Arg Phe Leu Leu Asn Val Ile Leu Val Leu Ala Ile Val Leu
Phe Leu Arg Tyr Val His Tyr Ser Leu Glu Pro Glu
            20
<210> 74
<211> 29
<212> PRT
<213> Artificial Sequence
<223> predicted twin-arginine signal peptides of B.
      subtilis
Met Phe Glu Ser Glu Ala Glu Leu Arg Arg Ile Arg Ile Ala Leu Val
Trp Ile Ala Val Phe Leu Leu Phe Gly Ala Cys Gly Asn
            20
<210> 75
<211> 37
<212> PRT
<213> Artificial Sequence
<223> predicted twin-arginine signal peptides of B.
      subtilis
Met Gln Lys Tyr Arg Arg Arg Asn Thr Val Ala Phe Thr Val Leu Ala
                                    10
Tyr Phe Thr Phe Phe Ala Gly Val Phe Leu Phe Ser Ile Gly Leu Tyr
                                25
                                                    30
Asn Ala Asp Asn Leu
        35
<210> 76
<211> 34
<212> PRT
<213> Artificial Sequence
<223> predicted twin-arginine signal peptides of B.
      subtilis
<400> 76
Met Met Leu Asn Met Ile Arg Arg Leu Leu Met Thr Cys Leu Phe Leu
                                    10
1
Leu Ala Phe Gly Thr Thr Phe Leu Ser Val Ser Gly Ile Glu Ala Lys
```

Asp Leu

```
<210> 77
<211> 44
<212> PRT
<213> Artificial Sequence
<220>
<223> predicted twin-arginine signal peptides of B.
      subtilis
<400> 77
Met Ala Glu Arg Val Arg Val Arg Lys Lys Lys Ser Lys
                                   10
Arg Arg Lys Ile Leu Lys Arg Ile Met Leu Leu Phe Ala Leu Ala Leu
                                25
Leu Val Val Val Gly Leu Gly Gly Tyr Lys Leu Tyr
                            40
<210> 78
<211> 47
<212> PRT
<213> Artificial Sequence
<220>
<223> predicted twin-arginine signal peptides of B.
      subtilis
<400> 78
Met Ser Asp Glu Gln Lys Lys Pro Glu Gln Ile His Arg Arg Asp Ile
Leu Lys Trp Gly Ala Met Ala Gly Ala Ala Val Ala Ile Gly Ala Ser
                               25
Gly Leu Gly Gly Leu Ala Pro Leu Val Gln Thr Ala Ala Lys Pro
                            40
<210> 79
<211> 54
<212> PRT
<213> Bacillus subtilis
<400> 79
Met Ala Tyr Asp Ser Arg Phe Asp Glu Trp Val Gln Lys Leu Lys Glu
1
Glu Ser Phe Gln Asn Asn Arg Phe Asp Arg Lys Phe Ile Gln Gly
                                25
Ala Gly Lys Ile Ala Gly Leu Ser Leu Gly Leu Thr Ile Ala Gln Ser
                            40
       35
Val Gly Ala Phe Glu Val
    50
<210> 80
<211> 65
<212> PRT
<213> Streptomyces coelicolor
Met Thr Pro Ala Asn His Gln Ala Pro Thr Ser Ala Pro Ser Pro Ala
                                    10
```

Pro Ser Gln Ser Ser His Ala Pro Glu Leu Arg Ala Ala Arg Ser Leu Gly Arg Arg Arg Phe Leu Thr Val Thr Gly Ala Ala Ala Leu 40 Ala Phe Ala Val Asn Leu Pro Ala Ala Gly Thr Ala Ser Ala Ala Glu Leu 65 <210> 81 <211> 60 <212> PRT <213> Streptomyces coelicolor <400> 81 Met Ala Pro Thr Gly Arg Pro Ser Ala Leu Ala Glu His Ala Phe Ser 10 Pro His Asp Ala Val Leu Gly Ala Ala Ala Arg His Leu Gly Arg Arg 25 Arg Phe Leu Thr Val Thr Ala Ala Ala Ala Leu Ala Phe Ser Thr 40 Asn Leu Pro Ala Arg Gly Ala Val Ala Ala Pro Glu <210> 82 <211> 47 <212> PRT <213> Streptomyces coelicolor <400> 82 Met Thr Ser Arg His Arg Ala Ser Glu Asn Ser Arg Thr Pro Ser Arg 10 Arg Thr Val Val Lys Ala Ala Ala Ala Gly Ala Val Leu Ala Ala Pro 25 Leu Ala Ala Ala Leu Pro Ala Gly Ala Ala Asp Ala Ala Pro Ala 40 <210> 83 <211> 53 <212> PRT <213> Streptomyces tendae <400> 83 Met Thr Pro Ala Ala Arg Pro Ser Gln His Ala Pro Glu Leu Arg Ala 5 10 Ala Ala Arg His Leu Gly Arg Arg Arg Phe Leu Thr Val Thr Gly Ala 25 Ala Ala Leu Ala Phe Ala Val Asn Leu Pro Ala Ala Gly Thr Ala 40 Ala Ala Glu Leu